

Claims

What is claimed is:

5 1. A method for identifying a polynucleotide sequence encoding a polypeptide of a wild ancestor or domesticated organism, wherein said polypeptide is or is suspected of being associated with enhanced economic productivity in said wild ancestor or domesticated organism, comprising the steps of:

10 a) comparing polypeptide-coding nucleotide sequences of said wild ancestor and said domesticated organism; and

15 b) selecting a polynucleotide sequence in either the domesticated organism or the wild ancestor that contains a nucleotide change as compared to the corresponding sequence in the wild ancestor or domesticated organism, respectively, wherein said change is evolutionarily neutral or positively evolutionarily significant, whereby the polynucleotide which encodes a polypeptide associated with enhanced economic productivity is identified.

20 2. A method for identifying a polynucleotide sequence encoding a polypeptide of a wild ancestor of a domesticated organism, wherein said polypeptide is or is suspected of being associated with a stress-resistance trait that is unique, enhanced or altered in the wild ancestor of the domesticated organism as compared to the domesticated organism, comprising the steps of:

25 a) comparing polypeptide-coding nucleotide sequences of said domesticated organism to polypeptide-coding nucleotide sequences of said wild ancestor; and

 b) selecting a polynucleotide sequence in the wild ancestor that contains a nucleotide change as compared to a corresponding sequence in the domesticated organism, wherein said change is evolutionarily neutral, whereby the wild ancestor's polynucleotide sequence is identified.

3. The method of claim 2 wherein said domesticated organism is a plant selected from the group consisting of maize, rice, tomato, potato and other domesticated plants whose ancestor is known.

4. The method of claim 3 wherein said domesticated plant is maize and said wild 5 ancestor is teosinte.

5. The method of claim 2 wherein the protein-coding nucleotide sequences of said domesticated organism correspond to cDNA.

6. The method of claim 2, wherein the nucleotide change is a non-synonymous substitution.

10 7. The method of claim 2, wherein the domesticated organism is a plant and the stress-resistance trait is selected from the group consisting of drought resistance, disease resistance, pest resistance, high salt level resistance and other stress-resistance traits of commercial interest.

15 8. A method of identifying an agent which may modulate a stress-resistance trait in a wild ancestor of a domesticated organism, said method comprising contacting at least one candidate agent with the wild ancestor, the domesticated organism, or with a cell or transgenic organism that expresses the polynucleotide sequence identified in claim 2, wherein the agent is identified by its ability to modulate the function of the polynucleotide or of the polypeptide encoded by the identified polynucleotide sequence.

20 9. A method for modulating stress-resistance in a wild ancestor of a domesticated organism by administering the agent of claim 8.

25 10. A method of identifying an agent to a polypeptide sequence encoded by a polynucleotide sequence in a domesticated organism that corresponds to the wild ancestor stress-resistance polynucleotide sequence of claim 2, comprising contacting at least one candidate agent with the domesticated organism, the ancestor organism, or with a cell or

transgenic organism that expresses the polynucleotide sequence, whereby the agent is identified by its ability to modulate function of the polypeptide sequence.

11. A method for modulating stress-resistance in a domesticated organism by administering the agent of claim 10.

5 12. A method for identifying an evolutionarily neutral change in a polypeptide-coding polynucleotide sequence of a wild ancestor of a domesticated organism comprising the steps of:

a) comparing polypeptide-coding polynucleotide sequences of said wild ancestor to corresponding sequences of said domesticated organism; and

10 b) selecting a polynucleotide sequence in said domesticated organism that contains a nucleotide change as compared to the corresponding sequence of the wild ancestor, wherein the change is evolutionarily neutral and the polynucleotide is associated with a stress-resistance trait in the wild ancestor of the domesticated organism, whereby the evolutionarily neutral change in the polynucleotide is identified.

15 13. The method of claim 12 wherein said domesticated organism is a plant selected from the group consisting of maize, rice, tomato, potato and other domesticated plants for which the ancestor is known.

14. The method of claim 12 wherein said domesticated plant is maize and said wild ancestor is teosinte.

20 15. The method of claim 12 wherein the protein-coding nucleotide sequences of said domesticated organism correspond to cDNA.

16. The method of claim 12, wherein the nucleotide change is a non-synonymous substitution.

25 17. The method of claim 12, where the domesticated organism is a plant and the relevant trait is selected from the group consisting of drought resistance, disease resistance,

pest resistance, high salt level resistance, and other stress resistance traits of commercial interest.

18. A method for large scale sequence comparison between polypeptide-coding nucleotide sequences of a wild ancestor of a domesticated organism and polypeptide-coding sequences from said domesticated organism, wherein the wild ancestor organism polypeptide confers or is suspected of conferring a stress-resistance trait that is unique, enhanced or altered in the wild ancestor as compared to the domesticated organism, comprising:

a) aligning the wild ancestor sequences with corresponding sequences from the domesticated organism according to sequence homology; and

10 b) identifying any nucleotide changes within the domesticated organism sequences or wild ancestor sequences as compared to the homologous sequences from the wild ancestor or domesticated organism, respectively.

15 19. The method of claim 18, wherein the domesticated organism is a plant selected from the group consisting of maize, rice, tomato and other domesticated plants whose ancestors are known.

20. The method of claim 19 wherein said domesticated plant is maize and said wild ancestor is teosinte.

21. The method of claim 18 wherein the protein-coding nucleotide sequences of said domesticated species correspond to cDNA.

20 22. A method for correlating an evolutionarily neutral nucleotide change to a stress-resistance trait that is unique, enhanced or altered in a wild ancestor of a domesticated organism comprising:

a) identifying a nucleotide sequence according to claim 2; and

25 b) analyzing the functional effect of the presence or absence of the identified sequence in the domesticated organism or ancestor organism.

23. A method for making a transfected plant cell or a transgenic plant comprising the steps of:

- a) transforming a plant cell to contain a polynucleotide encoding the stress-resistance polypeptide of a wild ancestor of claim 2, wherein said polynucleotide is operably linked to a promoter that can be used effectively for expression of transgenic proteins;
- 5 b) optionally growing and maintaining said cell under conditions whereby a transgenic plant is regenerated therefrom.

24. The transfected cell generated by the method of claim 23.

25. The transgenic plant generated by the method of claim 23.

10 26. A method for identifying a polynucleotide sequence encoding a polypeptide of a domesticated organism, wherein said polypeptide is or is suspected of being associated with improved yield in said domesticated organism as compared to a wild ancestor of said domesticated organism, comprising the steps of:

- a) comparing polypeptide-coding nucleotide sequences of said domesticated organism to polypeptide-coding nucleotide sequences of said wild ancestor; and
- 15 b) selecting a polynucleotide sequence in the domesticated organism that contains a nucleotide change as compared to a corresponding sequence in the wild ancestor, wherein said change is evolutionarily significant, whereby the domesticated organism's polynucleotide sequence is identified.

20 27. The method of claim 26 wherein said polypeptide that is associated with improved yield is an EG307 polypeptide.

28. A method for correlating an evolutionarily significant nucleotide change to a yield-related trait that is unique, enhanced or altered in a domesticated plant, comprising:

- a) identifying a nucleotide sequence according to claim 26; and
- 25 b) analyzing the functional effect of the presence or absence of the identified sequence in the domesticated plant.

29. A method for producing a transfected plant cell or transgenic plant comprising the steps of:

a) transfected a plant cell to contain a heterologous DNA segment encoding a polypeptide and derived from an EG307 polynucleotide not native to said cell; wherein said polynucleotide is operably linked to a promoter that can be used effectively for expression of transgenic proteins;

5 b) optionally growing and maintaining said cell under conditions whereby a transgenic plant is regenerated therefrom;

c) optionally growing said transgenic plant under conditions whereby said DNA is expressed, whereby the total amount of EG307 polypeptide in said plant is increased.

10 30. The method of claim 29, further comprising the step of obtaining and growing additional generations of descendants of said transgenic plant which comprise said heterologous DNA segment wherein said heterologous DNA segment is expressed.

31. Plant cells, comprising heterologous DNA encoding an EG307 polypeptide.

15 32. A propagation material of a transgenic plant comprising the transgenic plant cell according to claim 31.

33. A transgenic plant containing heterologous DNA which encodes an EG307 polypeptide that is expressed in plant tissue.

20 34. An isolated polynucleotide which includes a promoter operably linked to a polynucleotide that encodes the EG307 gene in plant tissue.

35. The isolated polynucleotide of Claim 34, wherein said polynucleotide is a recombinant polynucleotide.

36. The method of claim 34, wherein the promoter is the promoter native to an EG307 gene.

37. A method of making a transfected cell comprising:

- a) identifying an evolutionarily significant EG307 polynucleotide in an ancestor of a domesticated plant or a corresponding polynucleotide in a domesticated plant;
- b) using said EG307 polynucleotide to identify a non-polypeptide coding sequence that may be a transcription or translation regulatory element, enhancer, intron or other 5' or 3' flanking sequence;
- c) assembling a construct comprising said non-polypeptide coding sequence operably linked to a polynucleotide encoding a reporter protein; and
- d) transfecting said construct into a host cell.

10 38. A transfected cell produced according to the method of claim 37.

39. A method of making a transgenic plant comprising the method of claim 37, wherein said host cell is a plant cell, further comprising the step of growing and maintaining said cell under conditions whereby said transgenic plant is regenerated therefrom.

40. A transgenic plant produced by the method of claim 38.

15 41. A method of identifying an agent that modulates the function of the non-polypeptide coding regions of an evolutionarily significant EG307 polynucleotide, comprising contacting the transfected host cell of claim 37 with at least one candidate agent, wherein the agent is identified by its ability to modulate the transcription or translation of said reporter polynucleotide.

20 42. An agent identified by the method of claim 41.

43. A method of identifying an agent that modulates the function of the non-polypeptide coding regions of an evolutionarily significant EG307 polynucleotide, comprising contacting the transgenic plant of claim 39 with at least one candidate agent, wherein the agent is identified by its ability to modulate the transcription or translation of 25 said reporter polynucleotide.

44. An agent identified by the method of claim 43.

45. A transfected host cell comprising a host cell transfected with a construct comprising a promoter, enhancer or intron polynucleotide from an evolutionarily significant EG307 polynucleotide or any combination thereof, operably linked to a polynucleotide 5 encoding a reporter protein.

46. A method of identifying an agent which may modulate yield, said method comprising contacting at least one candidate agent with a plant or cell comprising an EG307 gene, wherein the agent is identified by its ability to modulate yield.

47. The method of Claim 46, wherein the plant or cell is transfected with a 10 polynucleotide encoding an EG307 gene.

48. An agent identified according to the method of claim 46.

49. The method of claim 46, wherein said identified agent modulates yield by modulating a function of the polynucleotide encoding the polypeptide.

50. The method of claim 46, wherein said identified agent modulates yield by 15 modulating a function of the polypeptide.

51. A method of providing increased yield in a plant comprising:

a) producing a transfected plant cell having heterologous DNA encoding an EG307 polypeptide whereby EG307 is expressed in said plant cell; and
b) growing a transgenic plant from the transfected plant cell wherein the EG307 20 transgene is expressed in the transgenic plant.

52. The method of claim 51, wherein the transgene is under the control of regulatory sequences suitable for controlled expression of the transgene.

53. A method of producing an EG307 polypeptide comprising:

a) providing a cell transfected with a polynucleotide encoding an EG307 polypeptide

positioned for expression in the cell;

- b) culturing the transfected cell under conditions for expressing the polynucleotide; and
- c) isolating the EG307 polypeptide.

5 54. A method of increasing yield in a plant comprising:

- a) producing a transgenic plant cell having a transgene containing the EG307 gene under the control of a promoter providing constitutive expression of the EG307 gene; and
- b) growing a transgenic plant from the transgenic plant cell wherein the EG307 transgene is expressed constitutively in the transgenic plant.

10 55. A method of providing controllable yield in a transgenic plant comprising:

- a) producing a transgenic plant cell having a transgene containing the EG307 gene under the control of a promoter providing controllable expression of the EG307 gene; and
- b) growing a transgenic plant from the transgenic plant cell wherein the EG307 transgene is controllably expressed in the transgenic plant.

15 56. The method of claim 55, wherein the EG307 gene is expressed using a tissue-specific or cell type-specific promoter, or using a promoter that is activated by the introduction of an external signal or agent, such as a chemical signal or agent.

57. A method of isolating a yield-related gene or fragment thereof from a plant cell, comprising:

- a) providing a sample of plant cell polynucleotides;
- b) providing a pair of oligonucleotides having sequence homology to a conserved region of an EG307 gene;
- c) combining the pair of oligonucleotides with the plant cell polynucleotides sample under conditions suitable for polymerase chain reaction-mediated polynucleotide amplification; and
- d) isolating the amplified yield-related polynucleotide or fragment thereof.

58. A plant yield-related polynucleotide isolated according to the method of claim
57.

59. A method of isolating a yield-related polynucleotide comprising:

5 a) providing a preparation of polynucleotides selected from the group consisting of

5 genomic plant cell DNA and recombinant plant cell library polynucleotides;

10 b) contacting the preparation with an EG307 oligonucleotide under hybridization
conditions providing detection of polynucleotides having 50% or greater sequence identity;
and

10 c) isolating a yield-related polynucleotide by its association with the EG307

oligonucleotide.

60. The method of Claim 59, wherein the EG307 oligonucleotide is detectably-labelled and the yield-related gene is isolated by its association with the detectable label.

61. The method of Claim 59, wherein the EG307 oligonucleotide is at least 12 nucleotides in length.

62. The method of Claim 59, wherein the EG307 oligonucleotide is at least 30 nucleotides in length.

63. A method of isolating a yield-related gene from plant cell polynucleotides comprising:

20 a) providing a sample of plant cell polynucleotides;

b) providing a pair of oligonucleotides having sequence homology to a conserved region of an EG307 gene;

c) combining the pair of oligonucleotides with the plant cell polynucleotides under conditions suitable for polymerase chain reaction-mediated polynucleotides amplification; and

25 d) isolating the amplified yield-related polynucleotide or fragment thereof.

64. A method of identifying a plant yield-related gene comprising:
a) providing a plant tissue sample;
b) introducing into the plant tissue sample a candidate plant yield-related gene;
c) expressing the candidate plant yield-related gene within the plant tissue sample;

5 and

d) determining whether the plant tissue sample exhibits change in yield response, whereby a change in response identifies a plant yield-related gene.

65. A plant yield-related gene isolated according to the method of Claim 64.

66. An isolated polynucleotide selected from the group consisting of:

10 a) a polynucleotide selected from the group consisting of SEQ ID NO:1, SEQ ID NO:91, SEQ ID. NO:2, SEQ ID NO:4, SEQ ID NO:5, SEQ ID NO:7, SEQ ID NO:10, SEQ ID NO:11, SEQ ID NO:12, SEQ ID NO:14, SEQ ID NO:15, and SEQ ID NO:17, and SEQ ID NO:18; and

15 b) a polynucleotide having at least 85% homology to a polynucleotide of a), and which confers substantially the same yield as the polynucleotide of a).

67. An isolated polynucleotide selected from the group consisting of:

20 a) a polynucleotide selected from the group consisting of SEQ ID NO:20, SEQ ID NO:21, SEQ ID. NO:23, SEQ ID NO:24, SEQ ID NO:25, SEQ ID NO:27, SEQ ID NO:28, SEQ ID NO:29, and SEQ ID NO:90; and

b) a polynucleotide having at least 85% homology to a polynucleotide of a), and which confers substantially the same yield as the polynucleotide of a).

68. An isolated polynucleotide selected from the group consisting of:

25 a) a polynucleotide selected from the group consisting of SEQ ID NO:33, SEQ ID NO:34, SEQ ID. NO:35, SEQ ID NO:37, SEQ ID NO:38, SEQ ID NO:40, SEQ ID NO:41, SEQ ID NO:42, SEQ ID NO:44, SEQ ID NO:45, SEQ ID NO:46, SEQ ID NO:47, SEQ ID NO:49, SEQ ID. NO:50, SEQ ID NO:51, SEQ ID NO:53, SEQ ID NO:54, SEQ ID NO:55, SEQ ID NO:57, SEQ ID NO:58, SEQ ID NO:60, SEQ ID NO:62, SEQ ID NO:63,

and SEQ ID NO:64; and

b) a polynucleotide having at least 85% homology to a polynucleotide of a), and which confers substantially the same yield as the polynucleotide of a).

69. An isolated polynucleotide selected from the group consisting of:

5 a) a polynucleotide selected from the group consisting of SEQ ID NO:66, SEQ ID NO:67, SEQ ID NO:69, SEQ ID. NO:70, SEQ ID NO:71, SEQ ID NO:73, SEQ ID NO:74, SEQ ID NO:75, SEQ ID NO:77, SEQ ID NO:59, and SEQ ID NO:78; and
b) a polynucleotide having at least 85% homology to a polynucleotide of a), and which confers substantially the same yield as the polynucleotide of a).

10 70. An isolated polynucleotide selected from the group consisting of:

a) a polynucleotide selected from the group consisting of SEQ ID NO:80, SEQ ID NO:81, and SEQ ID NO:82; and
b) a polynucleotide having at least 85% homology to a polynucleotide of a), and which confers substantially the same yield as the polynucleotide of a).

15 71. An isolated polynucleotide selected from the group consisting of:

a) a polynucleotide selected from the group consisting of SEQ ID NO:84 and SEQ ID NO:85; and
b) a polynucleotide having at least 85% homology to a polynucleotide of a), and which confers substantially the same yield as the polynucleotide of a).

20 72. An isolated polypeptide selected from the group consisting of:

a) a polypeptide encoded by a polynucleotide selected from the group consisting of SEQ ID NO:1, SEQ ID NO:91, SEQ ID. NO:2, SEQ ID NO:4, SEQ ID NO:5, SEQ ID NO:7, SEQ ID NO:10, SEQ ID NO:11, SEQ ID NO:12, SEQ ID NO:14, SEQ ID NO:15, and SEQ ID NO:17, and SEQ ID NO:18; and
b) a polypeptide encoded by a polynucleotide having at least 85% homology to a polynucleotide of a), and which confers substantially the same yield as the polynucleotide of a).

73. An isolated polypeptide selected from the group consisting of:
a) a polypeptide encoded by a polynucleotide selected from the group
consisting of SEQ ID NO:20, SEQ ID NO:21, SEQ ID. NO:23, SEQ ID NO:24, SEQ ID
NO:25, SEQ ID NO:27, SEQ ID NO:28, SEQ ID NO:29, and SEQ ID NO:90; and
5 b) a polypeptide encoded by a polynucleotide having at least 85% homology
to a polynucleotide of a), and which confers substantially the same yield as the
polynucleotide of a).

74. An isolated polypeptide selected from the group consisting of:
a) a polypeptide encoded by a polynucleotide selected from the group
consisting of SEQ ID NO:33, SEQ ID NO:34, SEQ ID. NO:35, SEQ ID NO:37, SEQ ID
NO:38, SEQ ID NO:40, SEQ ID NO:41, SEQ ID NO:42, SEQ ID NO:44, SEQ ID NO:45,
SEQ ID NO:46, SEQ ID NO:47, SEQ ID NO:49, SEQ ID. NO:50, SEQ ID NO:51, SEQ ID
NO:53, SEQ ID NO:54, SEQ ID NO:55, SEQ ID NO:57, SEQ ID NO:58, SEQ ID NO:60,
SEQ ID NO:62, SEQ ID NO:63, and SEQ ID NO:64; and
10 15 b) a polypeptide encoded by a polynucleotide having at least 85% homology
to a polynucleotide of a), and which confers substantially the same yield as the
polynucleotide of a).

75. An isolated polypeptide selected from the group consisting of:
a) a polypeptide encoded by a polynucleotide selected from the group
consisting of SEQ ID NO:80, SEQ ID NO:81, and SEQ ID NO:82; and
20 b) a polypeptide encoded by a polynucleotide having at least 85% homology
to a polynucleotide of a), and which confers substantially the same yield as the
polynucleotide of a).

76. An isolated polypeptide selected from the group consisting of:
a) a polypeptide encoded by a polynucleotide selected from the group
consisting of SEQ ID NO:84, SEQ ID NO:85; and
25 b) a polypeptide encoded by a polynucleotide having at least 85% homology

to a polynucleotide of a), and which confers substantially the same yield as the polynucleotide of a).

77. An isolated polypeptide selected from the group consisting of:

a) a polypeptide encoded by a polynucleotide selected from the group

5 consisting of SEQ ID NO:66, SEQ ID NO:67, SEQ ID NO:69, SEQ ID. NO:70, SEQ ID NO:71, SEQ ID NO:73, SEQ ID NO:74, SEQ ID NO:75, SEQ ID NO:77, SEQ ID NO:59, and SEQ ID NO:78; and

b) a polypeptide encoded by a polynucleotide having at least 85% homology to a polynucleotide of a).

10 78. An isolated polypeptide selected from the group consisting of:

a) a polypeptide encoded by a polynucleotide selected from the group

consisting of SEQ ID NO:84 and SEQ ID NO:85; and

b) a polypeptide encoded by a polynucleotide having at least 85% homology to a polynucleotide of a).